



Effects of Anthropogenic Dust Deposition on Lane Mountain Milkvetch (*Astragalus jaegerianus*)

Annual Report for Permit TE-022630-1



Prepared for:
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Ventura Fish and Wildlife Office
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U.S. DEPARTMENT OF THE INTERIOR
U.S. GEOLOGICAL SURVEY
WESTERN ECOLOGICAL RESEARCH CENTER

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2005

U.S. DEPARTMENT OF THE INTERIOR
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U.S. GEOLOGICAL SURVEY
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Introduction

Many factors threaten the survival of Mojave Desert plant species. One of the most prevalent and far-reaching is human expansion and development, which not only impacts species directly through plant injury and habitat loss but also generates many indirect threats as well (Belnap and Warren 2002; Lovich and Bainbridge 1999). Atmospheric dust generated by soil surface disturbance is one indirect threat that is particularly relevant for the federally endangered Lane Mountain Milkvetch (*Astragalus jaegerianus*).

Lane Mountain Milkvetch (*Astragalus jaegerianus*) has a limited range in the western Mojave Desert. A large portion of this habitat is located within the proposed expansion area for the US Army National Training Center (NTC). Remaining habitat for the Lane Mountain Milkvetch is found on Bureau of Land Management (BLM) land south of the NTC. The US Fish and Wildlife Service has listed the Lane Mountain Milkvetch as endangered due to its endemism and its sensitivity to military and other vehicular traffic (Federal Register 55:6184-6229 and Federal Register 61:7596-7613). A better understanding of the impact from proposed military uses will be useful in evaluating prospects for long-term conservation and recovery of this species.

Plans are underway to fence certain populations to prevent direct damage by military vehicles (Federal Register 69: 18018-18035), but concern remains that habitat quality within these protected areas will be adversely affected by airborne dust raised by training maneuvers. Long term exposure to a degraded habitat could counteract the effectiveness of the primary means of protection. Studying the effects of dust deposition on Lane Mountain Milkvetch's physiology, growth and reproduction is necessary for assessing the feasibility of protecting populations that occur along high activity roads.

In dry regions like the American southwest, eolian dust is a natural occurrence. Dust storms in the western part of the Mojave Desert are primarily generated by cyclonic activity during winter and spring (Brazel and Nickling 1987). Dry lakebeds, or playas, are typical within the region where Lane Mountain Milkvetch is found, providing a source of fine particulate dust. In addition, human activity on desert soils such as military transports and training maneuvers greatly increases fugitive dust, resulting in suspension of particles in the atmosphere. Similarly, portions of the desert denuded of vegetation by development or off-highway vehicles (OHV) produce exposed areas from which dust can be raised (Adams et al. 1982). Human induced changes in playa hydrology may also increase nutrient fluxes and

influence plant communities that are downwind (Blank et al. 1999).

In a review, Farmer (1993) reported that leaf surface dust deposition may affect photosynthesis, respiration, transpiration and allow penetration of phytotoxic gaseous pollutants. Sharifi et al. (1997) demonstrated that Mojave Desert perennials proximate to military activities at the NTC exhibit a 21-58% reduction in photosynthesis as well as total shoot length. If leaf surface dust reduces photosynthesis in the Lane Mountain Milkvetch, over time decreased growth and reproduction could reduce the persistence of this rare species.

The goal of this study was to determine whether surface dust accumulation could impede the persistence of Lane Mountain Milkvetch. We hypothesized that photosynthesis would decrease with accumulation of dust on the leaves, and would consequently inhibit growth and reproduction. In addition, we examined the site characteristics that influence the amount of airborne dust that can be intercepted by the plant canopy.

Methods

Field

In April 2004, we selected 20 Lane Mountain Milkvetch plants from the previously surveyed Coolgardie Mesa population located on land administered by California BLM. The plants were randomly assigned one of five dust concentration treatments: 0, 8, 16, 24 and 32 g/m², with four replicate plants per concentration (total n = 20). Beginning in early May 2004, we dusted the leaf canopies of the study plants at these levels biweekly until plants senesced at the end of June. In comparison to our dust treatments, heavily dusted *Atriplex canescens* growing near tank trails at the NTC had dust concentrations of 40 g dust m⁻² leaf area during summer drought (Sharifi et al. 1997).

Dust was generated by collecting soil from nearby roadbeds and passing it through a 45 µm sieve (24% of dust was < 20 µm). This procedure produced dust particles small enough to fall through the L-shaped trichomes and onto the surface of the leaf. We distributed a pre-measured quantity of dust onto the plant through a 45 µm sieve to ensure uniformity of application.

We chose three shoots of approximately equal size for monitoring growth and reproduction for each plant. Shoot and leaf length, leaflet number, and flower/fruit production were recorded biweekly throughout the study period (May 7 through June 21). During the same sampling periods, we measured leaf-level net photosynthesis (P_{net}) for each plant using an open, compensating photosynthesis system (Li-6400, Li-Cor,

Inc., Lincoln, NE). We chose the newest, fully formed leaf from each plant to measure photosynthesis. The same leaf was then excised, and midday water potentials (ψ) were measured with a Scholander-type pressure chamber (PMS Instrument Co., Corvallis, OR). Photosynthesis measurements were taken between 9:00 am and 12:00 pm, and midday water potentials were measured immediately between noon and 1:00 pm. We verified leaf dust concentration in two ways: 1) excised leaves used for physiology measurements were soaked in de-ionized water for 24 hours and 2) an additional leaf (adjacent to the leaf used for physiological measurements) was rinsed with 20 mL of de-ionized water. The resulting residue was dried to a constant mass in a convection oven at 50°C and weighed. It was determined later that the physiology leaves overestimated the dust concentration because cell solubles leaked from the leaves and inflated the mass of the residue. Therefore, we used the adjacent leaves for our estimates of leaf dust concentration throughout the experiment. Leaf dust concentration and net photosynthesis were expressed on a leaf-area basis by recording an image of each leaf with light-sensitive diazo paper and measuring the area using a leaf-area meter (Li-3000A, Li-Cor Inc.).

To determine the levels of ambient dust deposition for plants in the Coolgardie population, we constructed dust traps based on a design by Marith Reheis (1995), but we did not include the metal straps often used to discourage perching birds. Traps were installed at varying distances from one and two-track vehicular routes. Fifteen traps were placed within the Coolgardie population, with 4 traps placed among the study plants and 11 associated with additional milkvetch plants. The traps were positioned on fence posts at shrub height, roughly 3-4 feet above the soil surface. Traps were rinsed out monthly using de-ionized water, and the residue dried and weighed.

Greenhouse

We established 25 Lane Mountain Milkvetch individuals in the greenhouse in February 2004 with seeds collected from the Goldstone population in 2003. The seeds were scarified using sandpaper (R. Sharifi and B. Prigge, pers. comm.) to ensure germination. Seeds were planted in pots modified from 30 cm length PVC pipe (10 cm diameter) using wire mesh on the bottom to allow for drainage. Shoots were supported with trellises fashioned from plastic chicken wire and later transplanted to 5 gal pots to discourage constriction of the root system which might have influenced photosynthetic responses.

In an attempt to elucidate results obtained from the field experiment, we dusted plants in the greenhouse beginning December 16, 2004 and ending January 27, 2005. Of the original 25 plants, 10 were healthy

enough to include in the experiment. Plants were randomly assigned either a dusted treatment or non-dusted control ($n = 5$ per treatment). Two shoots of equivalent size were chosen to monitor growth. Dust was created and applied weekly using the field protocol. We determined dust concentration by rinsing leaves with 20 mL of de-ionized water. The treated plants were visibly dusty compared to the untreated control plants and retained dust on the shoots and leaves between applications. At the end of each month, we washed three leaves of each target plant to determine cumulative dust concentration. Shoot length, leaf number and number of branching points were recorded weekly on the growth shoots. Leaf-level net photosynthesis was also measured weekly on one "new" leaf and one "old" leaf from each plant. New leaves were characterized as the most recent, fully formed leaf on a shoot. Old leaves were initially selected by tagging the fifth leaf from the growing tip on a randomly chosen shoot. Once the initial leaf showed signs of senescence, like yellowing or desiccation, we switched to the next younger leaf on the same shoot.

Analysis

Experimental manipulation of dust concentration in the field was challenging because of the difficulty of dusting whole canopies of disparate branching architecture, as well as varied wind direction and velocity between sampling periods (Fig. 1). Though we were unable to maintain target dust concentrations, we did apply a constant pressure of dust throughout the study period and achieved a dust concentration gradient.

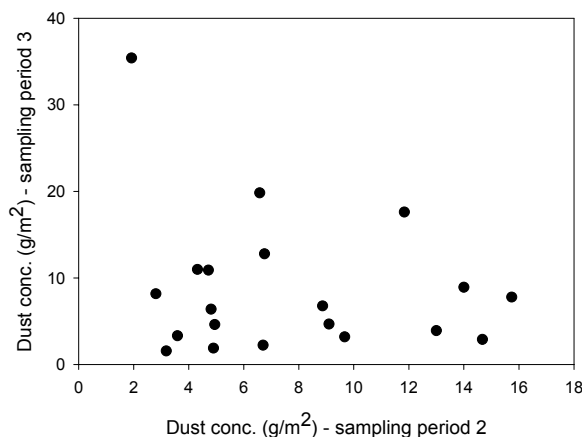


Fig 1. Dust concentrations measured on leaves two weeks after application did not reflect dust concentrations originally applied ($P = 0.37$, $F_{1,18} = 0.84$, $R^2 = 0.04$), indicating low retention of dust between applications.

To express this constant pressure, we integrated the fluctuating dust concentration over the study period, using the trapezoidal rule (SigmaPlot, v. 8.0,

Richmond, CA) to obtain the average dust concentration that an individual plant experienced over the duration of the study. Photosynthesis and xylem water potential were similarly expressed as an integrated average using the trapezoidal rule. Physiology data from the fourth sampling period were omitted because plants had begun senescing by this time.

All data were analyzed using SAS statistical software (v. 9.1, Cary, NC). Patterns in plant physiology and growth associated with dust applied to plants in the field were analyzed using simple linear regression. One individual was omitted from analyses because it began senescing soon after the first dust application. Growth and gas-exchange responses to dust treatments applied to greenhouse plants were analyzed using nonparametric Wilcoxon rank sum test and two-factor (dust treatment and leaf age) split-plot ANOVA, respectively. A multiple linear regression was used to determine whether dust trap characteristics (elevation and distance from single- and dual-track vehicle routes), maximum and minimum air temperatures and rainfall could predict dust trap concentrations (backward elimination, $\alpha = 0.10$).

Results

Field

Phenological measurements indicated a steady increase in twig length across all plants until most plants reached peak growth in mid June. Twig elongation was reduced at higher dust concentrations (Fig. 2), but the number of leaves each plant produced over the study period did not change significantly (Fig. 3).

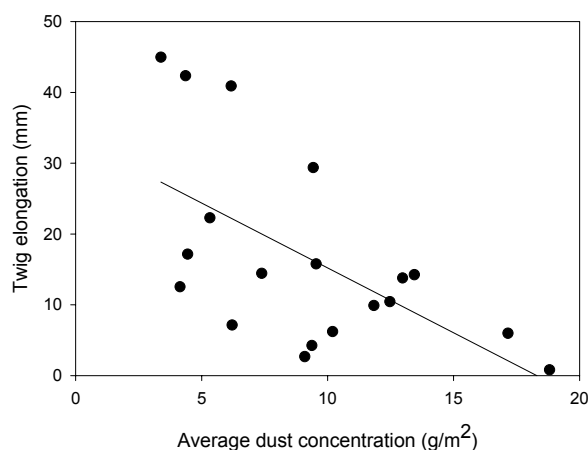


Fig. 2. Twig elongation (i.e., increase in twig length through the season) declined with increasing leaf dust concentration ($P < 0.01$, $F_{1,18} = 9.41$, $R^2 = 0.36$). Twig elongation (mm) = $33.52 - 1.83 \times \text{dust concentration (g/m}^2\text{)}$.

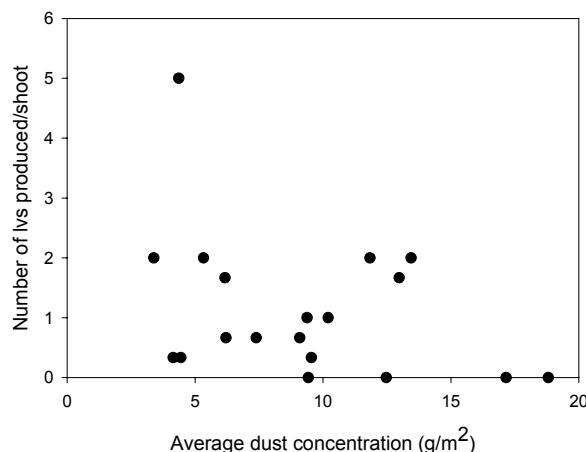


Fig. 3. Leaf production did not change significantly as leaf dust concentration increased ($P = 0.12$, $F_{1,18} = 2.61$, $R^2 = 0.13$).

We do not report effects of dust on reproduction because plants initiated reproduction before dust treatments were applied (i.e., flower buds and developed flowers were present on twigs marked for growth and whole plants, respectively), and many of the flowers we originally observed were aborted.

Net photosynthesis increased as the concentration of dust on leaves increased (Fig. 4). This result contradicted our original hypothesis and is counter-intuitive because twig elongation decreased at higher dust concentrations. We subsequently hypothesized that plants that were heavily dusted allocated resources away from twig growth toward producing new leaves, which typically have higher photosynthetic rates. Therefore, heavily dusted plants with greater leaf production would have the highest photosynthesis rates averaged over the whole plant. This hypothesis was tested in the greenhouse by quantifying leaf production as well as measuring photosynthesis on both “old” and “new” leaves for each treatment (see results for “Greenhouse” below).

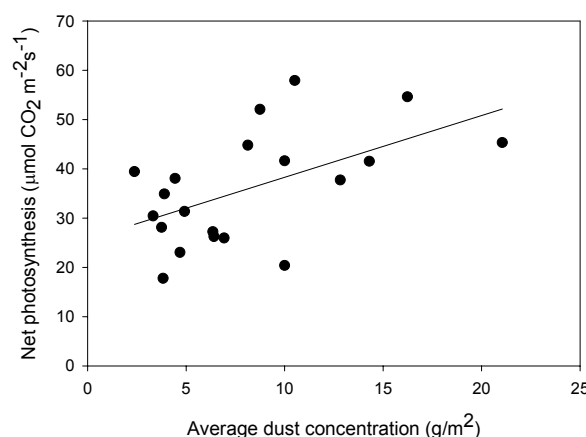


Fig. 4. Average leaf-level net photosynthesis increased with greater leaf dust concentrations ($P = 0.01$, $F_{1,18} = 7.51$, $R^2 = 0.29$).

The milkvetch plants did not show any difference in water stress across the dust concentration gradient (Fig. 5), and were within the range previously reported for this species (Gibson et al., 1998).

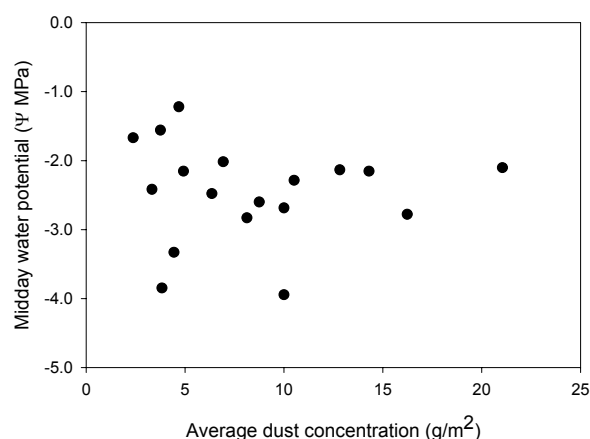


Fig. 5. Average midday water potentials did not change significantly with increasing leaf dust concentration ($P = 0.86$, $F_{1,17} = 0.03$, $R^2 = 0.00$).

Dust accumulation in the traps (May - December 2004) ranged from 10 to 20 g/m^2 (Fig. 6). The study area is intersected by one unimproved single-track and several dual-track vehicular routes, and we tested whether distance of traps from these routes as well as rainfall, maximum and minimum air temperatures and elevation influenced dust deposition.

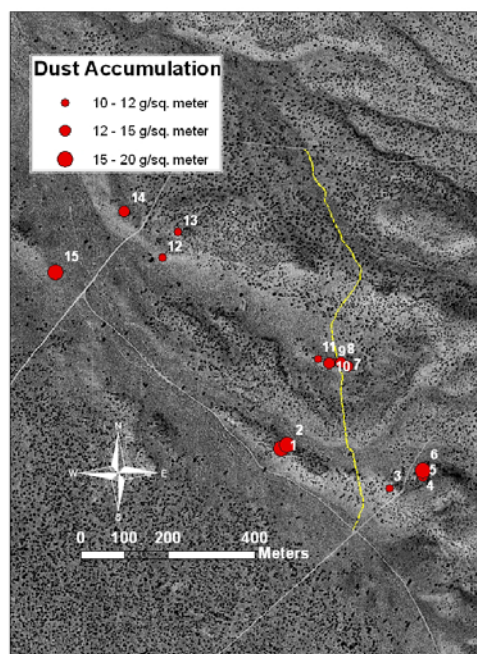


Fig. 6. Traps #12-15 are located among the study plants while the remaining traps are associated with additional Lane Mountain Milkvetch plants. A single-track route is highlighted in yellow; the rest are dual-track routes. (Image collected in 1998)

Rainfall and distance to dual-track routes were significant factors explaining dust concentration (overall model, $P < 0.01$, $F_{2,116} = 39.96$, $R^2 = 0.41$). Highest dust concentrations were associated with lower monthly rainfall (Fig. 7) and greater proximity to dual track roads. Wind direction and speed are likely to vary throughout the area as well but were not recorded for this study. Interestingly, dust collected in July and August, following heavy rains, was visibly oily and difficult to remove from traps. Contaminated dust has been observed in other traps located around the periphery of the Los Angeles Basin and is likely due to smog (M. Reheis, pers. comm.), but it remains to be seen whether this “greasy” dust has a detrimental effect on the milkvetch’s physiology and growth.

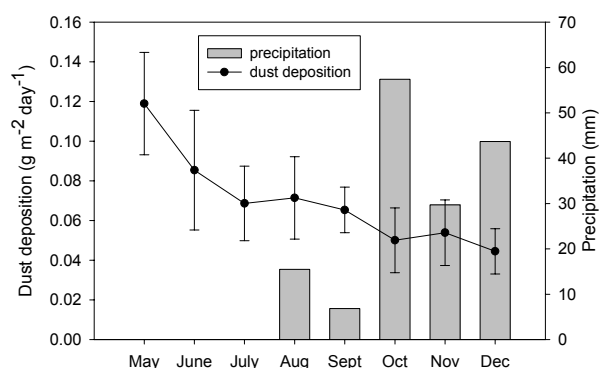


Fig. 7. Mean ambient dust deposition (± 1 s.d.) collected in traps over the study area was highest during the late spring months when milkvetch were active. No rain fell during May, June and July.

Greenhouse

Cumulative dust concentration at the end of the experiment for the treated plants remained between 20-40 g/m^2 . Although patterns in growth mirrored those in the field (dusted plants with lower twig elongation but little difference in leaf production), we found no statistically significant difference between dusted and control plants for either response (Fig. 8).

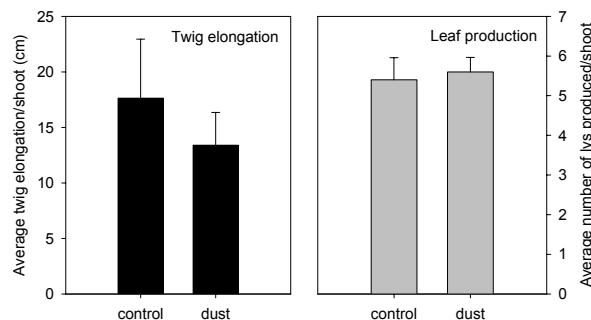


Fig. 8. Twig elongation (± 1 s.d.) and leaf production were measured on greenhouse grown plants ($n = 5$ for each dust treatment). Neither twig elongation ($P = 0.38$, $z = 0.31$) nor leaf production ($P = 0.12$, $z = 1.163$) were significantly different between the two greenhouse treatments.

Likewise, while dusted plants appear to have higher seasonal photosynthesis than controls (Fig. 9), this difference was not statistically significant. New leaves, however, had significantly higher rates of photosynthesis than old leaves.

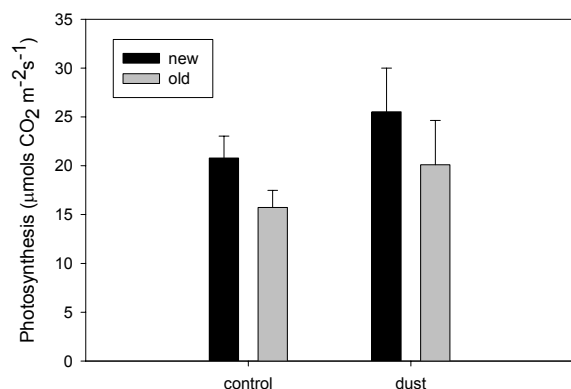


Fig. 9. Net photosynthesis was not different between the control and dusted treatments ($P = 0.33$, $F_{1,8} = 1.06$). However, new leaves did exhibit significantly higher rates of photosynthesis compared to old leaves ($P = 0.04$, $F_{1,8} = 5.76$).

Discussion

Studies of dust deposition on desert plants suggest that photosynthesis and growth are negatively impacted by the dust that falls on the surfaces of leaves (Beatley 1965; Sharifi et al. 1997, 1999). Even though some of the dust that was experimentally applied to Lane Mountain milkvetch plants was not retained on the leaf surfaces of plants in this field study (Fig. 1), the net accumulation of dust that did remain after repeated applications significantly reduced twig growth. Milkvetch are rarely ever observed growing outside of the canopy of desert shrubs (Prigge et al. 2000) which they use as a trellis for support (Gibson et al. 1998). In addition, milkvetch require at least two-thirds full sunlight to reach maximum net photosynthetic rates (Gibson et al. 1998). Consequently, we expect that milkvetch experiencing prolonged exposure to dust next to heavily used vehicle routes may not attain the height needed to acquire sunlight to achieve maximum net photosynthetic rates. Flower and fruit production during the study was poor irrespective of dusting treatment, thus it remains to be seen whether reproductive effort over the life of the plant is compromised by diminished growth.

Although dusted milkvetch plants in the field had lower twig growth, average net photosynthesis was higher as leaf dust concentrations increased, which was contrary to our original hypothesis. We subsequently dusted plants in the greenhouse to determine whether higher net photosynthesis reflected the production of new leaves, which would lead to enhanced photosynthesis averaged over the whole plant. Similar

to the milkvetch studied in the field, plants dusted in the greenhouse tended to have lower twig elongation, greater net photosynthesis, and produced the same number of new leaves compared to non-dusted plants, though the results were not statistically significant. While it is possible that we were unable to detect a significant treatment difference due to the low number of plants available for the greenhouse experiment, enhanced water status of greenhouse plants may have counteracted the negative effects of dust. Sharifi et al. (1999) showed that naturally dusted *Larrea tridentata* plants that received irrigation had higher shoot water potentials and more growth than non-irrigated dusted plants. The greenhouse milkvetch plants were watered regularly, which may have been sufficient to negate any effect of dust accumulation. While we have inadequate evidence to conclude that greater net photosynthesis in dusted plants is a consequence of greater leaf production, alternatively the dusted milkvetch plants may reallocate nitrogen to the photosynthetic apparatus to ensure adequate carbon uptake under stressful conditions. This trade-off between nitrogen allocation to photosynthesis and to structural/support tissues may explain why shoot growth was diminished while photosynthesis was elevated (Onoda et al. 2004; Takashima et al. 2004).

Our results indicate that for every gram per square meter increase in leaf dust concentration there was a 1.83 mm decrease in shoot elongation. Ambient levels of dust deposition for the Coolgardie population during May (1.66 g/m^2) and June (1.20 g/m^2), as extrapolated from daily rates determined by dust traps, were below the lowest level of dust that was applied biweekly during the field experiment (8 g/m^2). Therefore, we expect that milkvetch plants associated with unimproved vehicle routes for this population at the current use will not be greatly affected by the dust they receive (all of the study plants have recovered from experimental dusting and put out new growth for the 2005 season). However, future protection of this population will depend on minimizing use of dual-track routes, especially during periods of low rainfall when dust evolution is greatest. Moreover, for populations located in the expansion area of the NTC, and where fugitive dust is expected to increase due to increased military training, proposed fencing of population segments will likely benefit if an adequate buffer area between the dust source and the plants is implemented.

Recommendations

Dust generated by recreation and expanded military training in milkvetch habitat is a concern for the continued persistence of plants, whether protected in conservation areas or in areas that are fenced to prohibit direct losses of individuals. Low ambient dust levels for the Coolgardie population during this study

do not currently pose a threat to that population. However, dust traps should remain in the area to monitor dust generation as increased recreational use of the area is a concern. For low to medium intensity training areas expected within the NTC expansion area, dust impacts can be reduced by providing sufficient distance between the source of dust and the populations. In order to design appropriate access around existing milkvetch populations, and for subsequent effectiveness monitoring, additional information is required:

- 1) The amount of dust generated from training areas adjacent to milkvetch populations should be characterized spatially using dust traps positioned at varying distances and orientations from dust sources. This characterization should begin prior to expansion of military training so that appropriate buffer zones can be determined. Current heavy use routes can serve as a proxy for routes expected to occur following expansion by placing traps at increasing distances and varying orientations from where milkvetch plants would occur relative to expected vehicular use in the expansion area. Monthly monitoring of dust, in conjunction with rainfall measurements gathered from accompanying rain gauges, will continue to be important in understanding the seasonal nature of climate, dust generation and milkvetch phenology.
- 2) The amount of dust that is retained on leaves after it falls on a milkvetch canopy remains a difficult variable to quantify due to differences in canopy architecture, host shrub associations, and variable wind speeds and durations throughout the growing season. However, this measure is essential to understanding prolonged dust impacts to milkvetch physiology, growth and reproduction. Dust collected in traps positioned next to existing milkvetch plants should be evaluated as an accurate correlate of actual leaf dust concentrations, and this relationship could be adequately tested using other species in current training areas as proxies before expansion occurs.
- 3) The level of leaf dust concentration that significantly reduces milkvetch reproduction should be determined to define buffer zones between milkvetch populations and the low to medium intensity training areas. This target reproductive rate for milkvetch should consider the reproductive output required to maintain minimum viable populations and is currently being investigated by USGS scientists.
- 4) Monitoring of dust deposition as well as measurements of milkvetch physiology, growth and reproduction should be in place after expansion occurs so that efficacy of buffers can be assessed

and adjusted to reduce pressure on existing populations.

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